AMENDMENTS TO THE SPECIFICATION

Please amend the paragraphs: [Title], [0010], [0011], [0015], [0044], [0045], [0047], [0048], [0049], [0058], [Table 2], [Table 3], [Table 4], [0097], [00110], [00111], [00112], [Table 7], [Table 8], [00113], [00114], [00115], [00128], [00129], [00133], [00134], [00136], [00141], [00142], [00143], [00144], and [Abstract], as follow:

[Title] ~~CRYSTALLIZATION OF~~ WILD-TYPE KINASE DOMAIN OF HUMAN EPHRIN RECEPTOR A2 (EPHA2) AND CRYSTALLIZATION THEREOF

[0010] In one variation, the protein has activity characteristic of EPHA2. For example, the protein may optionally be inhibited by inhibitors of wild type EPHA2. The protein crystal may also diffract X-rays for a determination of structure coordinates to a resolution having a value of 4Å, 3.5Å, 3.0Å or less.

[0011] In one variation, the protein crystal has a crystal lattice in a [[P3221]] $P3_221$ space group. The protein crystal may also have a crystal lattice having unit cell dimensions, +/- 5%, of a=72.12Å, b= 72.12Å and c=241.62Å.

[0015] The method may optionally further comprise forming a protein crystal that has a crystal lattice in a [[P3221]] $P3_221$ space group. The method also optionally further comprises forming a protein crystal that has a crystal lattice having unit cell dimensions, +/- 5%, of a=72.12Å, b= 72.12Å and c=241.62Å. The invention also relates to protein crystals formed by these methods.

[0044] Figure [[3A]] 3 lists ~~a first set of~~ the atomic structure coordinates for EPHA2 (reference numbers in Col. E corresponding to the residue numbering for SEQ ID NO: 1) as derived by X-ray crystallography from a crystal that comprises the protein which is present as a dimer. Drawing sheets 3-3AO of Figure 3 list the structure coordinates of chain A; drawing sheets 3AP-3CD of Figure 3 list the structure coordinates of chain B. The following

abbreviations are used in Figure 3: "X, Y, Z" crystallographically define the atomic position of the element measured; "B" is a thermal factor that measures movement of the atom around its atomic center; "Occ" is an occupancy factor that refers to the fraction of the molecules in which each atom occupies the position specified by the coordinates (a value of "1" indicates that each atom has the same conformation, i.e., the same position, in all molecules of the crystal).

[0045] delete

[0047] Figure 5A illustrates AMP-PNP bound in the active site of EPHA2 based on the determined crystal structure for the molecule in the asymmetric unit corresponding to the structure coordinates of chain A shown in ~~Figure 3A.~~ drawing sheets 3-3AO of Figure 3.

[0048] Figure 5B illustrates AMP-PNP bound in the active site of EPHA2 based on the determined crystal structure for the molecule in the asymmetric unit corresponding to the structure coordinates of chain B shown in ~~Figure 3A.~~ drawing sheets 3AP-3CD of Figure 3.

[0049] Figure 6 illustrates a system that may be used to carry out instructions for displaying a crystal structure of [[EphA2]]EPHA2 encoded on a storage medium.

[0058] In another embodiment, EPHA2 comprises of SEQ. ID No. 3 which comprises the portion of the kinase domain of wild-type EPHA2 that are represented in both the first and second sets of structure coordinates shown in ~~Figures 3A and 3B~~ Figure 3 (reference numbers in Col. E of Figure 3 refer to residue numbering for SEQ ID NO: 1).

[Table 2]

Table 2: ATP binding site residues within 4 Angstroms of the AMP-PNP binding site (SEQ ID NO: 1).

ILE 619	VAL 627	GLU 693
GLY 620	ALA 644	TYR 694
ALA 621	LYS 646	MET 695

GLY 622	THR 692	LEU 746
GLU 623		

[Table 3]

Table 3: ATP binding site residues within 7 Angstroms of the AMP-PNP binding site (SEQ ID No. 1).

ILE 619	ALA 644	ASN 697
GLY 620	ILE 645	GLY 698
ALA 621	LYS 646	ALA 699
GLY 622	GLU 663	LYS 702
GLU 623	ILE 676	ARG 743
PHE 624	ARG 677	ASN 744
GLY 625	THR 692	LEU 746
VAL 627	GLU 693	VAL 747
TYR 628	TYR 694	SER 756
LYS 629	MET 695	ASP 757
VAL 643	GLU 696	

[Table 4]

Table 4: ATP binding site residues within 10 Angstroms of the AMP-PNP binding site (SEQ ID No. 1).

LYS 617	PHE 660	LYS 702
VAL 618	GLU 663	PHE 703
ILE 619	MET 667	ARG 705
GLY 620	ILE 675	GLU 706
ALA 621	ILE 676	ASP 739
GLY 622	ARG 677	ALA 741
GLU 623	LEU 678	ALA 742
PHE 624	GLU 679	ARG 743
GLY 625	ILE 690	ASN 744
GLU 626	ILE 691	ILE 745
VAL 627	THR 692	LEU 746
TYR 628	GLU 693	VAL 747
LYS 629	TYR 694	ASN 748
GLY 630	MET 695	SER 749
PRO 642	GLU 696	LYS 754
VAL 643	ASN 697	VAL 755
ALA 644	GLY 698	SER 756
ILE 645	ALA 699	ASP 757

LYS 646	LEU 700	PHE 758
THR 647	ASP 701	GLY 759
LYS 778		

[0097] EPHA2 crystals also preferably are capable of diffracting X-rays for determination of atomic coordinates to a resolution having a value of 4 Å, 3 Å, 2.5 Å, 2 Å or ~~greater~~ less.

[00110] Each unit cell comprised two EPHA2-AMP-PNP-$(Mg^{2+})_2$ complexes. Structure coordinates were determined for each of the two complexes and the resultant two sets of structure coordinates from the refinement are presented in ~~Figures 3A and 3B~~ Figure 3.

[00111] It is noted that the sequence of the structure coordinates of chain A and chain B presented in ~~Figures 3A and 3B~~ Figure 3 differ in some regards from the sequence shown in SEQ. ID No. 1.

[00112] For some residues, the electron density obtained was insufficient to identify the side chain. As a result, the side chains of these residues were truncated such that a different amino acid is reported. Tables 6 and 7 summarize the differences between SEQ. ID No. 1 and the truncated residues of chain A and chain B, appearing in ~~Figures 3A and 3B~~ drawing sheets 3-3AO and 3AP-3CP of Figure 3, respectively.

[Table 7]

Table 7: Truncated Residues in The Structure Coordinates of ~~Figure 3A~~ Chain A (Figure 3) (SEQ ID NO: 1).

T605-A605	K633-A633	T634-A634
K638-A638	K639-A639	L760-A760
K793-A793	K828-A828	R858-A858
R876-A876	F887-A887	

[Table 8]

Table 8: Truncated Residues in The Structure Coordinates of ~~Figure 3B~~ Chain A (Figure 3) (SEQ ID NO: 1).

L602-A602	K633-A633	K649-A649
E654-A654	Q656-A656	R657-A657
K684-A684	K686-A686	L764-A764
K778-A778	K793-A793	K828-A828
M840-A840	R860-A860	K882-A882
T883-A883		

[00113] It is also noted that structure coordinates are not reported for some residues because the electron density obtained was insufficient to identify the position of these residues. For ~~Figure 3A~~ chain A, structure coordinates for residues 596-604, 635-637, 761-777 and 888-895 are not reported. For ~~Figure 3B~~ chain B, structure coordinates for residues 596-601, 635-638, 765-777, 884-895 are not reported.

[00114] Those of skill in the art understand that a set of structure coordinates (such as those in Figure ~~3A and 3B~~ 3) for a protein or a protein-complex or a portion thereof, is a relative set of points that define a shape in three dimensions. Thus, it is possible that an entirely different set of structure coordinates could define a similar or identical shape. Moreover, slight variations in the individual coordinates may have little effect on overall shape. In terms of binding pockets, these variations would not be expected to significantly alter the nature of ligands that could associate with those pockets. The term "binding pocket as used herein refers to a region of the protein that, as a result of its shape, favorably associates with a ligand

[00115] These variations in coordinates may be generated because of mathematical manipulations of the EPHA2 structure coordinates. For example, the sets of structure coordinates shown in Figure ~~3A and 3B~~ 3 could be manipulated by crystallographic permutations of the structure coordinates, fractionalization of the structure coordinates,

application of a rotation matrix, integer additions or subtractions to sets of the structure coordinates, inversion of the structure coordinates or any combination of the above.

[00128] The three-dimensional crystal structure of EPHA2 may be generated, as is known in the art, from the structure coordinates shown in ~~Figures 3A or 3B~~ Figure 3 and similar such coordinates.

[00129] The refined crystal structure of EPHA2-AMP-PNP determined according to the present invention contains amino acids residues 605-887 as numbered according to SEQ. ID No. 1 (based on the coordinates of ~~Figure 3A~~ chain A), one bound AMP-PNP molecule, and two Mg^{2+} ions. A total of 66 water molecules were included.

[00133] Figure 5A illustrates AMP-PNP bound in the active site of EPHA2 based on the determined crystal structure for the molecule in the asymmetric unit corresponding to the structure coordinates of chain A shown in ~~Figure 3A~~ drawing sheets 3-3AP of Figure 3. As can be seen, the salt bridge between conserved K646 and E663 is not maintained in the structure.

[00134] Figure 5B illustrates AMP-PNP bound in the active site of EPHA2 based on the determined crystal structure for the molecule in the asymmetric unit corresponding to the structure coordinates of chain B shown in ~~Figure 3B~~ drawing sheets 3AO-3CD of Figure 3. As can be seen, the salt bridge between conserved K646 and E663 is maintained in the structure.

[00136] The term "binding site" or "binding pocket", as the terms are used herein, refers to a region of a protein that, as a result of its shape, favorably associates with a ligand or substrate. The term "EPHA2-like binding pocket" refers to a portion of a molecule or molecular complex whose shape is sufficiently similar to the EPHA2 binding pockets as to bind common ligands. This commonality of shape may be quantitatively defined by a root mean square deviation (rmsd) from the structure coordinates of the backbone atoms of the amino acids that make up the binding pockets in EPHA2 (as set forth in ~~Figure 3A or 3B~~ Figure 3).

[00141] With the knowledge of the EPHA2 crystal structure provided herein, Applicants define an EPHA2 binding pocket as a binding pocket where the relative positioning of the 4, 7, and/or 10 Angstroms sets of amino acids are substantially conserved. Again, it is noted that it may be desirable to form variants where 1, 2, 3, 4 or more of the residues set forth in Tables 2, 3, and 4 are varied in order to evaluate the roles these amino acids play in the binding pocket. Accordingly, any set of structure coordinates for a protein from any source having a root mean square deviation of non-hydrogen atoms of less than 2 Å when superimposed on the non-hydrogen atom positions of the corresponding atomic coordinates of either ~~Figure 3A or Figure 3B~~ chain A or chain B for the 4, 7, and/or 10 Angstroms sets of amino acids shall be considered identical. As noted previously, the root mean square deviation is intended to be limited to only those non-hydrogen atoms of amino acid residues that are common to both the protein ~~fragment~~ fragments represented in ~~Figures 3A or 3B~~ Figure 3 and the protein whose structure coordinates are being compared to the coordinates shown in ~~Figures 3A or 3B~~ Figure 3 since the sequence of the protein may be varied somewhat.

[00142] Accordingly, in one embodiment, the invention relates to data, computer readable media comprising data, and uses of the data where the data comprises the structure coordinates of either chain A or chain B shown in ~~Figures 3A or 3B~~ Figure 3 or structure coordinates having a root mean square deviation of non-hydrogen atoms of less than 2 Å when superimposed on the non-hydrogen atom positions of the corresponding atomic coordinates of either chain A or chain B in Figure 3 ~~Figures 3A or 3B~~ for the 4, 7, and/or 10 Angstroms sets of amino acids.

[00143] Again, it is noted that the root mean square deviation is intended to be limited to only those non-hydrogen atoms of amino acid residues that are common to both the protein fragment represented in one or more of the tables and the protein whose structure coordinates are being compared to the coordinates shown in ~~Figure 3A or 3B~~ Figure 3.

[00144] As noted above, there are many different ways to express the surface contours of the EPHA2 structure other than by using the structure coordinates provided in ~~Figure 3A or 3B~~

Figure 3. Accordingly, it is noted that the present invention is also directed to any data, computer readable media comprising data, and uses of the data where the data defines a computer model for a protein binding pocket, at least a portion of the computer model having a surface contour that has a root mean square deviation of less than 3 Å when superimposed on a surface contour defined by atomic coordinates of ~~Figure 3A or 3B~~ Figure 3, the root mean square deviation being calculated based only on non-hydrogen atoms in the structure coordinates of ~~Figure 3A or 3B~~ Figure 3 that are present in residues shown in Tables 2, 3, and/or 4.

[Abstract] Provided are crystals relating to human Ephrin Receptor A2 and its various uses.